U.S. DEPARTMENT OF COMMERCE
National Technical Information Service

PB-256 698

Distribution of Ingested Americium in Chickens and Transport to Eggs

Environmental Monitoring & Support Lab.

May 1976

PB 256 698

REPRODUCED BY
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U.S. DEPARTMENT OF COMMERCE
SPRINGFIELD, VA. 22161

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TECHNICAL REPORT DATA
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1. REPORT NO. EPA-600/3-76-058	2.
3. RECIPIENT'S ACCESSION NO.	
4. TITLE AND SUBTITLE DISTRIBUTION OF INGESTED AMERICIUM IN CHICKENS AND TRANSPORT TO EGGS	5. REPORT DATE May 1976
	6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S) Anita A. Mullen, Stephen R. Lloyd, and Robert E. Mosley	8. PERFORMING ORGANIZATION REPORT NO.
9. PERFORMING ORGANIZATION NAME AND ADDRESS Environmental Monitoring and Support Laboratory-LV U.S. Environmental Protection Agency P. O. Box 15027 Las Vegas, NV 89114	10. PROGRAM ELEMENT NO. 1FA628
	11. CONTRACT/GRANT NO.
12. SPONSORING AGENCY NAME AND ADDRESS Same as above	13. TYPE OF REPORT AND PERIOD COVERED Final FY75
	14. SPONSORING AGENCY CODE EPA-ORD Office of Health & Ecological Effects

15. SUPPLEMENTARY NOTES

16. ABSTRACT

The soluble citrate complex of americium-241 was orally administered to 20 white Leghorn laying hens daily for 2 weeks. The yolks, whites, and shells from the eggs were analyzed for their americium content. Yolk was the only egg fraction in which radioactivity was observed. The americium-241 activity in yolks reached a maximum on the 14th day of dosing. Biological half-times of 2.00 ± 0.18 days and greater than 33 days were indicated by the average concentration values of americium-241 in yolks laid after the maximum activity was reached.

The hens were serially sacrificed at 1, 10, and 20 days after the final administration of americium-241. Tissue samples were collected and the americium content determined in the edible portions and feathers of the hens. Americium was detected in most tissues shortly after dosing; the main concentrations were found in the liver and the skeleton. The highest concentration per organ (3.03×10^{-3} percent of the dose) occurred in the liver of the hens sacrificed 10 days after final administration of americium-241.

PRICES SUBJECT TO CHANGE

KEY WORDS AND DOCUMENT ANALYSIS

17. a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Americium Chickens Citrates Eggs Radioactive Isotopes Radiation Dosage	Americium in Chickens Citrate Complex Distribution and Transport Exposure/Dose Assessment	06C 06R 07B 07C 07E 18B

18. DISTRIBUTION STATEMENT RELEASE TO PUBLIC	19. SECURITY CLASS (This Report) UNCLASSIFIED	21. NO. OF PAGES 15
	20. SECURITY CLASS (This page) UNCLASSIFIED	22. PRICE $2.50 - 2.25

EPA Form 2220-1 (9-73)

☆GPO 691-220-1978

EPA-600/3-76-058
May 1976

DISTRIBUTION OF INGESTED AMERICIUM IN CHICKENS AND TRANSPORT TO EGGS

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INTRODUCTION

The necessity for obtaining further information concerning the fate of the transuranic nuclides in the environment has been well established. However, little is known about the metabolism of americium in chickens *(Gallus domesticus)* and its subsequent transfer to eggs. In the event that farm areas become contaminated with americium, chickens allowed to forage around such farmyards may ingest contaminated soil and feedstuffs. This ingested americium may be metabolized and transferred to egg contents and edible body tissues, thereby causing contamination of a portion of the human food chain.

The absorption, distribution, and excretion of the actinide elements in biological systems are determined largely by their chemical characteristics, which are similar to those of the lanthanides (Durbin, 1973; Hamilton, 1948). Lanthanide fission products administered intramuscularly to laying hens were found to be distributed throughout the tissues with a major fraction concentrated in the skeleton and a lesser fraction in the liver (Mraz et al., 1964). Of the egg components, the yolk was found to contain amounts ranging from 18.1 percent of the administered dose of lanthanum-140 to 1.8 percent of the administered dose of praseodymium-142 per four-egg composite sample.

Several investigators have found indications that plutonium and americium may be metabolized in a manner similar to iron (Lindenbaum and Rosenthal, 1972; Taylor, 1972). Transferrin appears to be the universal transporting protein for plutonium (IV) in the blood plasma of all animals that have been studied. The main route of elimination of plutonium is through the gastrointestinal tract, via the bile and feces, which is also similar to that for iron. Laying hens require a large amount of iron in addition to their maintenance need, since the average egg yolk contains 1.1 mg of this element (Maynard and Loosli, 1962). If plutonium and americium are metabolized in a manner similar to that reported for iron, then the egg yolk should contain these elements.

This study was conducted to determine the extent to which the relatively soluble americium citrate is absorbed and distributed in the edible tissues and egg components after oral administration to laying hens.

SUMMARY

Americium is one of the transuranic elements which, as a daughter product of the radioactive decay of plutonium, may be found in the environment as a result of worldwide fallout or accidental contamination. One of the routes to man is through the food chain. This study investigated the transport through one portion of the food chain, chickens and eggs. The soluble citrate complex of americium-241 was orally administered to 20 white Leghorn laying hens daily for 2 weeks. The yolks, whites, and shells from the eggs were analyzed for their americium content. Yolk was the only egg fraction in which radioactivity was observed. The americium-241 activity in yolks reached a

maximum on the 14th day of dosing indicating that equilibrium had not been reached and additional transfer of americium to yolk would have occurred if dosing had continued. Analysis of egg yolk yielded a two component excretion curve with biological half-times of 2.00 ± 0.18 days and greater than 33 days, indicated by the average concentration values of americium-241 in yolks laid after the maximum activity was reached.

The hens were serially sacrificed at 1, 10, and 20 days after the final administration of americium-241. Tissue samples were collected and the americium content determined in the edible portions and feathers of the hens. Americium was detected in most tissues at early times with the main concentration in the liver and skeleton. The highest concentration per organ (3.03×10^{-3} percent of the dose) occurred in the liver of hens sacrificed 10 days after final administration of americium-241.

In a contaminated farmyard where chickens are raised with equal likelihood of consuming equivalent proportions of plutonium and americium, the americium would present the greater hazard to those persons eating eggs and/or chicken liver from that source.

CONCLUSIONS

Ingestion of the relatively soluble citrate form of americium-241 by laying hens resulted in the contamination of the egg yolks and the concentration of americium-241 in the skeletons and livers of the hens. The activity per egg yolk continued to increase during the 14 days of americium ingestion, indicating that equilibrium had not occurred and that americium concentration may increase in yolks for a much longer period of time. It appears that more americium-241 concentrates in egg yolk and chicken liver than plutonium-238 when ingested in the relatively soluble citrate form. This concentration should be recognized when contaminating events occur in the vicinity of a farming area.

RECOMMENDATIONS

Although little hazard to humans would be expected from ingesting americium-241 contaminated eggs or chicken livers, the possibility of this contamination must be recognized in the event of the contamination of farming areas. Further study would determine the concentration of ingested americium oxide, the most probable form of released americium. The biological availability of the organically incorporated americium should be investigated. Knowing the genetic effects of americium on chicks hatched from contaminated eggs and the concentrations of americium in these chicks and its transfer ability to their eggs after they reach maturity would be of additional help in evaluating the hazard of americium in the environment.

EXPERIMENTAL PROCEDURES

Thirty white Leghorn laying hens were housed in stainless steel animal metabolism cages with a center partition arranged to allow each hen to have an individual cage area. Excreta were collected in paper-lined trays beneath

the wire mesh. The cages were cleaned daily to decrease airborne contamination and to keep the eggs clean.

The chickens were acclimated to their surroundings for several weeks prior to the study. During the study, the chickens were maintained on commercial laying pellets and had access to water and oyster shell.

The chickens used in this study were beginning their second laying cycle and were about 2 years of age.

SOURCE PREPARATION

The solution for dosing was prepared fresh daily from americium-241 (III) nitrate and citrate buffer (pH 3.5 to preserve the monomeric state) and will be referred to as americium citrate. This preparation allows better comparison with previous experiments in which plutonium solutions were prepared similarly.

DOSING

Each of the 20 hens was administered 2 microcuries of americium-241 citrate orally by means of a dosing needle and syringe every day for 2 weeks.

Ten control hens were caged randomly among the dosed chickens. The control hens did not receive a placebo, but were used only as biological samplers to measure any cross-contamination.

SAMPLE COLLECTION AND STORAGE

Eggs were collected daily. After washing they were broken and the whites, yolks, and shells were separated. The separated fractions were weighed, placed in counting containers, and refrigerated until analyzed.

Six hens were sacrificed at 1 day, seven hens at 10 days, and seven hens 20 days after final dose administration. After sacrifice and exsanquination, tissue samples consisting of gizzard, heart, liver, and muscle were collected to determine the amount of americium present in the edible tissues. Feathers were collected to ascertain if the concentration of americium occurred therein. The feathers were washed with a commercial decontamination solution placed in an ultrasonic generator prior to analysis to remove accumulated surface contamination.

SAMPLE ANALYSIS

The counting containers were covered with plastic film and turned film-side down on the thin window crystal detector of a FIDLER instrument. The detector was housed in a standard shield and coupled to a RIDL single-channel analyzer system with scaler readout. This system was calibrated for various sample types and weights. Sample collection was stopped when the counting error exceeded 30 percent.

RESULTS AND DISCUSSION

In order to follow the possible mechanisms for absorption of americium by chickens and its subsequent transfer to eggs, it is necessary to review some of the physiology of digestion and egg production. The following information is excerpted from P. D. Sturkie's (1970) treatise on the subject.

Digestive organs of chickens are similar to those of mammals except that birds have a gizzard and a crop. The crop is formed as a dilatation of the esophagus and is a storage organ. The proventriculus of the bird is comparable to the simple glandular stomach of mammals. The gizzard is a highly specialized organ for grinding, as the bird has no teeth.

Rate of passage of food through the alimentary canal is influenced by the consistency, hardness, and water content of the food and the amount consumed. If the crop is full, it requires 10 to 18 hours for all of the food to leave the crop. In experiments with dye-impregnated food, it was indicated that the dye can be detected in the feces within 24 hours of administration. When radioactive barium was administered to chickens, it appeared that approximately half the activity was excreted in 4 to 5 hours.

Although information about digestion is meager, the evidence indicates that little digestion occurs in the mouth or the crop. Enzymes contained in pure intestinal juice have not been determined, but mixed intestinal juice contains pancreatic juice and proteases in an alkaline medium.

The pH of the contents of digestive organs in the chicken is: crop, 4.5; proventriculus, 4.4; gizzard, 2.6; duodenum, 5.7-6.01; jejunum, 5.8-5.9; ileum, 6.3-6.4; rectum or colon, 6.3; cecum, 5.7. In live birds, the large intestine was slightly alkaline (7.1). The acidity of avian bile (5.88 in chickens) may, in part, account for the lower pH in the alimentary tract of birds as compared to mammals. The secretion rate of bile in white Leghorn chickens is about 1 milliter per hour, or about 9.5 milliter/kilogram of body weight per day. The role of bile in avian digestion has not been fully studied, but it is presumed that it aids in the absorption of fats by its emulsifying action and activating effects on pancreatic lipases. Little fat is absorbed until the third segment of small intestine is reached, where 42 to 60 percent (depending on type) is absorbed. Most of the remaining fat (90 to 98 percent) is absorbed in the fourth segment. Little fat is absorbed in the cecum or large intestine.

The liver has many functions other than its digestive action. There are pronounced sex differences in blood levels of plasma proteins, lipids, and calcium of birds. These differences are due primarily to the effect of the female sex hormone on the liver in the formation and retention of these substances. The clearance rate of liver, as measured by its ability to extract sodium bromsulphthalein (BSP) from blood and excrete it in the bile, is markedly influenced by estrogen and, as expected, the clearance rate is greater in laying hens.

The development of an egg starts in the ovary, where the yolk portion is formed. Here there are many ova, each enclosed in a follicle. The yolk is deposited in concentric layers during growth and maturation, and these rings may vary in color, depending on the amount of yellow pigment, or xanthophyll,

in the food. When the process is complete, the follicle bursts and the yolk, surrounded by a membrane, passes into the oviduct. This process may take from 7 to 10 days. As the yolk passes into the magnum, the albumin of the egg is secreted around the ova. This process takes approximately 3 hours; then the egg passes into the isthmus, where the inner and outer shell membranes are formed in about an hour and a quarter, before the water is added through the semi-permeable shell membrane to the albumin and where the egg shell is deposited. The egg remains in the shell gland approximately 18 to 20 hours, then passes to the vagina for laying (P. D. Sturkie's, 1970).

There are many problems inherent in the interpretation of the data derived from studies of this nature. Not only do the hens vary in the length of time they take to produce an egg, but the production of eggs is variable, with some hens laying an egg a day for weeks while others lay sporadically. Thus, a large range of activity can be anticipated in an egg laid at any given time after ingestion of a radionuclide. Considering the above variables, the range of the data shown in Table 1 would be expected.

TABLE 1. TRANSFER OF ORALLY ADMINISTERED AMERICIUM TO EGG YOLK

	Total Dose (μCi)	Time to Peak (days)	Peak Activity Range (pCi/18-g yolk)	Peak Activity Average (pCi/18-g yolk)	Half-time ($T_{\frac{1}{2}}$ days)
^{241}Am	26.7	14	60-2,900	870	(1) 2.00±0.18 (2) >33

The transfer of americium to the egg yolks is presented in Figure 1. Each point represents the average activity per yolk in eggs laid during that time period. The americium activity increased over the dosing period with the maximum, 0.00326 percent of the total administered activity, occurring in eggs laid after the fourteenth dose. Since nc obvious plateau was evident, additional doses might have resulted in a higher activity. The americium activity in the egg yolks decreased with a half-time of 2.00 ± 0.18 days followed by a second component of greater than 33 days. Americium was not detected in the white or shell of the eggs, nor in any portion of the eggs from the control hens.

These results would appear to agree in general with published data on the metabolism of iron. In an investigation to determine the metabolism of manganese and iron by poultry (Panic, 1970), it was determined that measurable quantities of iron-59 were found only in the egg yolk. An iron-59 concentration of 1.90 percent of the dose per yolk appeared in eggs laid 51 hours after oral administration, with the peak concentration of 2.80 percent per yolk occurring 6.1 days after initial ingestion. It was found that iron is transported from the blood to the yolk by a protein or proteins deposited in yolks only. Furthermore, the iron is more firmly bound to the protein phosvitin than to transferrin. Phosvitin appears in hen plasma during the laying period and is deposited only in the yolk.

Figure 1. Average concentration of americium-241 in egg yolks from chickens ingesting 2 microcuries/day for 14 days

Americium was absorbed by the hens with tissue concentrations occurring mainly in the liver and skeleton (see Table 2). These results agree with those of Taylor (1962) studying the metabolism of americium in rats.

In previous studies conducted with plutonium, it was found that chickens absorbed a very small percentage of plutonium-238 ingested as the relatively insoluble dioxide. This small percentage of absorption and transfer to egg yolk appears to present little hazard to humans consuming the eggs. Laying hens absorbed a somewhat higher percentage of the nuclide after ingesting a more soluble form of plutonium-238 than that absorbed from ingestion of the dioxide-form. Also, a large percentage of the absorbed plutonium was transferred to the yolk of the eggs. In older laying hens ingesting either form of plutonium-238, plutonium does not appear to concentrate in tissue in amounts large enough to constitute a hazard when human consumption is limited to normal quantities. This may not be true of the livers of younger poultry raised solely for food although little hazard could be expected.

TABLE 2. ACTIVITY IN TISSUE FROM CHICKENS INGESTING 2 μCi AMERICIUM-241 CITRATE EACH DAY FOR 14 DAYS (AVERAGE AND STANDARD DEVIATION)

Tissue	Days After Final Ingestion	% Dose/g	Mean Weight Total Organ, g	% Dose/organ
Gizzard w/o lining	1 (6)*	$9.08\ 2.96\times10^{-6}$	24.9	2.26×10^{-4}
	10 (7)	$8.67\ 4.45\times10^{-6}$	21.7	1.88×10^{-4}
	20(7)	$2.48\ 1.71\times10^{-6}$	21.6	5.30×10^{-5}
Liver	1	$3.36\ 3.11\times10^{-5}$	41.9	1.41×10^{-3}
	10	$8.00\ 2.28\times10^{-5}$	37.9	3.03×10^{-3}
	20	$7.47\ 8.58\times10^{-6}$	42.4	3.16×10^{-4}
Gall Bladder w/bile	1	$4.27\ 2.21\times10^{-6}$	2.6	1.11×10^{-4}
	10	$6.53\ 9.28\times10^{-6}$	2.0	1.31×10^{-5}
	20	$6.85\ 2.60\times10^{-6}$	1.0	7.20×10^{-6}
Heart	1	$4.27\ 0.76\times10^{-6}$	8.4	3.58×10^{-5}
	10	$8.73\ 8.67\times10^{-6}$	6.6	5.76×10^{-5}
	20	$5.68\ 3.80\times10^{-6}$	6.2	3.55×10^{-5}
Muscle	1	$1.28\ 0.50\times10^{-6}$		
	10	$7.01\ 3.80\times10^{-7}$		
	20	$6.59\ 2.29\times10^{-7}$		
Fat	1	$3.23\ 3.02\times10^{-6}$		
	10	$3.53\ 1.43\times10^{-6}$		
	20	$2.63\ 2.69\times10^{-6}$		
Long bone	1	$1.94\ 1.15\times10^{-5}$		
	10	$8.35\ 6.26\times10^{-6}$		
	20	$4.19\ 5.47\times10^{-6}$		
Sternum	1	$7.49\ 2.74\times10^{-6}$		
	10	$1.26\ 0.76\times10^{-5}$		
	20	$6.42\ 6.93\times10^{-6}$		
Rib	1	$8.80\ 4.27\times10^{-6}$		
	10	$1.98\ 2.55\times10^{-5}$		
	20	$5.77\ 5.63\times10^{-6}$		
Blood	1	$7.00\ 5.39\times10^{-6}$		
	10	$4.19\ 5.43\times10^{-6}$		
	20	$1.05\ 0.46\times10^{-6}$		

*Number of samples.

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